



**REPORT ON A VISIT TO GUNUNG MADU PLANTATIONS
(EAST SUMATRA)
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INTRODUCTION AND GENERAL CONTEXT

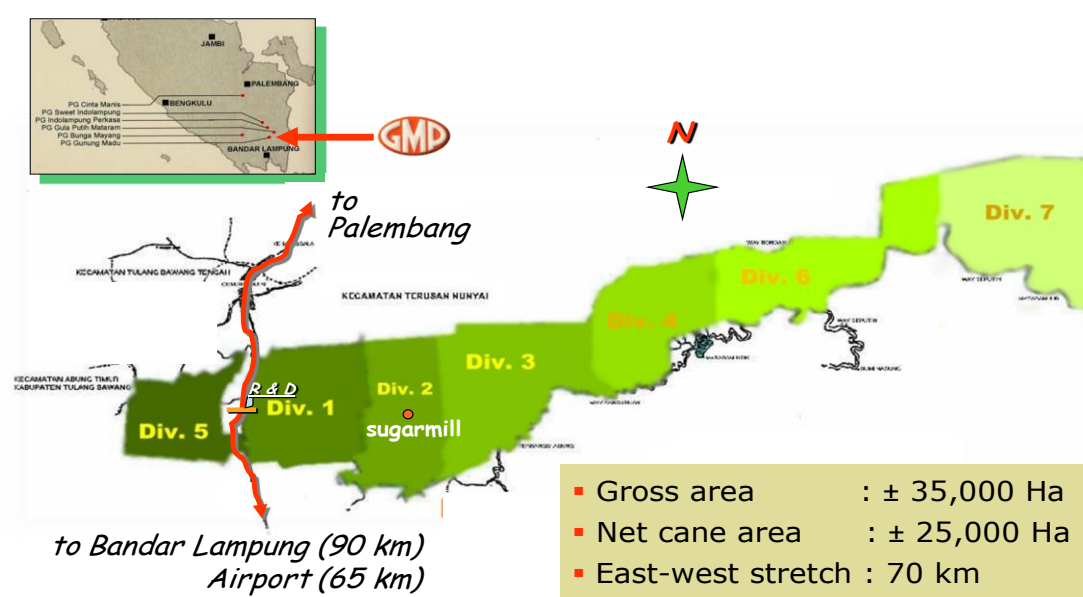
First of all, I would particularly like to thank Saefudin Saeroji for the invitation and the perfect organization of this visit to Gunung Madu Plantation (GMP). I also extend my thanks to the research directors and staff that made my stay very comfortable and useful. The main objective of this visit was to review the current biological control and other pest management strategies (including all necessary documents) based on field and laboratory visits and give some advice and recommendations to improve the technology and methods currently in place for a better pest management at GMP. As per our agreement with the research department we have focused our visit and recommendations on *Trichogramma chilonis* and *Cotesia flavipes*, the most effective parasitoids. At the end of the report we propose some useful experiments that can easily be implemented at GMP.

1. BRIEF OVERVIEW OF GMP (see map)

Gunung Madu Plantations has been established in 1975 and is located in South Sumatra (Lampung province), at 70 kms from its capital Lampung. Data on the production are the following:

- It's an integrated sugar plantation (cane plantation and sugar factory)
- The total land area is 35,000 Ha, out of which 25,000 Ha is under sugarcane crop for milling
- The factory's milling capacity is 14,000 TCD (ton canes per day)
- The harvesting & milling season is from April to early November (2010 season is the 34th season)
- The sugar production is around 180,000 tons/season
- Varieties (8 to 10) are mainly from GMP (60%).

• *Map of PT Gunung Madu Plantations*



2. THE SUGARCANE MOTH BORERS AND PARASITIDS IN USE AT GMP

Like any other producing countries, stalk borers are key pests of sugarcane. The spotted stemborer (*Chilo sacchariphagus*), the glossy stemborer (*Chilo auricilius*), and the top borer (identified as *Scirpophaga nivella intacta*, a name that need to be confirmed and compared with the other species present in Java *Scirpophaga excerptalis*). Based on the last 10 years data, damage levels are from 4.75 to 11.66% for the stem-borer (internode bored), and 4.37 to 10.03% for the top-borer (% dead-hearts or % infested tillage).

A general survey on the main varieties from 2007/08 to 2010/11 has shown that the infestation level has increased from year to year and today this level is above the international standard level of 5% of internodes borers (stemborers). The level of 10% of stalk damaged in 2010/2011 by *Scirpophaga* spp. is also a warning for pest managers. To effectively control moth borers, biological control and varietal resistance are the main components of IPM widely recommended and used by many sugarcane research institutes and factories. GMP has been using biological control against moth borers since the 80s.

Other pest problems at GMP are presented in the table below.

Common name	Species	Order : Family	Infested parts
White grub	<i>Pachnessa nicobarica</i> <i>Phyllophaga bidentata</i>	Coleoptera : Scarabaeidae	Root
Woolly aphid	<i>Ceratovacuna lanigera</i>	Hemiptera : Aphididae	Leaf
Migratory locust	<i>Locusta migratoria</i>	Orthoptera : Acrididae	Leaf
Rat	<i>Rattus</i> spp.	Rodentia: Muridae	Stem
Pink mealybug	<i>Saccharicoccus sacchari</i>	Homoptera : Pseudococcidae	Stem, young shoot & young root
Grey borer	<i>Eucosma schistaceana</i>	Lepidoptera : Eucosmidae	Bud
Leafhopper	<i>Perkinsiella saccharicida</i>	Homoptera : Delphacidae	Leaf vein
Mite	<i>Oligonychus exsicicator</i>	Acarina : Tetranychidae	Leaf blade

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Biological control at GMP

GMP has clearly made the choice of a biocontrol strategy and an interesting production of natural enemies has been developed and is implemented in the field all year round. However it is rather difficult to measure the impact of this strategy in the field as no clear assessment has been undertaken so far. In addition, it seems that since 2008 the borer infestation is increasing and it is therefore essential to understand the key drivers of such situation. There is also a real need to assess the efficacy of the releases of natural enemies in sugarcane fields/blocks.

These concerns are shared by the research department at GMP and hence explain the need to analyze the forces and weaknesses of biocontrol methods and tools in use. Here are some useful data given by the research department:

Parasitoid reared and released:

Trichogramma chilonis (egg parasitoid of stem-borers), *Cotesia flavipes* (larval parasitoid of *C. sacchariphagus*), *Sturmiopsis inferens* (larval parasitoid of *C. auricilius*), and *Elasmus zehntneri* (larval parasitoid of top-borer) are the natural enemies bred at the research department laboratories.

Production capacity of the rearing laboratories:

- *T. chilonis* : 300 millions/month
- *C. flavipes* : 1.5 millions/month
- *S. inferens* : 2,000/month
- *E. zehntneri* : 40,000/month

Rate of application and frequency of release:

- *T. chilonis* : 14,000/ha, once a month, up to 10-month old canes
- *C. flavipes* : 280/ha, on 6-month old canes
- *S. inferens* : 15 females/ha, on 6-month old canes
- *E. zehntneri* : 200/ha, on 6-month old canes

Number of people working in our biocontrol program: 60 casual workers, 11 permanent workers/employees, 1 entomologist.

3. REVIEW OF THE BIOLOGICAL CONTROL PROGRAM AND MAIN RECOMMENDATIONS

As said earlier, this review and recommendations are mainly for *Trichogramma* and *Cotesia* and their use in biological control.

3.1. Rearing of *Corcyra cephalonica*, the rice moth and *T. chilonis*:

This host is widely used in tropical sugar countries to produce *Trichogramma* spp., particularly in India. It is easy to breed and can give good results when all rearing conditions are in place.

Following our visit to this laboratory rearing which consists of small rooms dedicated to this species, the conclusion is that *Corcyra* moths are not produced in optimal conditions. Several problems have been identified:

- The development of another insect, *Tribolium* spp. (Coleoptera) which competes with *Corcyra* larvae and slow down the production
- The high temperature (35 to 40°C during the visit) due to the absence of a basic air-conditioning device and the confinement of the room.
- The absence of standard humidity (70-80°C)
- The absence of a light regime.
- The darkness and the cleanness of the rooms

- These negative points clearly impede the research department to establish a satisfactory production in terms of the quality of the insects: reduction of insect weight, mating rate, longevity and the fecundity (number of egg produced).

The management should therefore look carefully into these negative points and try to solve them one by one. They are clearly not insurmountable and we think that easy and non expansive solutions can be set up to improve the overall rearing conditions. For example we have made the following suggestions to the crop protection team and the management:

- The problem of *Tribolium* spp. can be reduced by putting pheromone traps. The pheromone lures can be purchased either in India (a company has to be selected) or UK (Greenwich University, the email address can be provided any time).
- The high temperature can be simply lowered and stabilized by installing an air-conditioning split system with a remote control.
- The Humidity can be controlled by a dehumidifier/humidifier device that is easy to find on the market.
- The rooms need clearly to be repainted in white or yellow and equipped with an aerating system (equipped with small propellers).
- The rooms should be equipped with proper lights that can be switch off and on by a remote control programmer.
- It would be interesting to explore the possibility of vacuum cleaning the moths recently emerged (normally with no damage). In fact the production of *Corcyra* could be slightly mechanized which will also reduce the time spent on manual operations. Lastly it is always useful to review some papers on moth rearing methods and follow the recommended procedures. Some Indian papers are really useful to improve the rearing conditions and the quality control of the production.

Parasitoids of *Trichogramma chilonis* are produced on *Corcyra* eggs (paper cards) in fairly good conditions. Each card normally contains 2500 parasitized eggs and is maintained in laboratory for incubation until the eggs turn black which mean they are ready to be sent to the fields. However, we have also noticed that there is no air conditioning in the storage room and the management should pay attention to this and provide the necessary optimal conditions as described above.

We are convinced that the improvement of the rearing conditions for *Corcyra cephalonica* will be a major boost for the production at GMP, not only for this host but for *Trichogramma*. The research department has the capacity to fill the challenge.

3.2. Field releases of *T. chilonis*

Trichogramma spp. (Hymenoptera: Chalcididae: Trichogrammatidae) are extremely tiny egg parasitoids widely used on sugarcane and other crops in the world to control

moth borers of economic importance. They are characterized by wings covered by hairs layered in radiant lines. Once mass-produced and released, the tiny parasites seek out and destroy eggs of caterpillar pests, such as sugarcane borers, codling moths, cotton bollworms, corn borers, spruce budworms and many others. The result is a living, biological 'insecticide' that strikes only the target pest with no risk to other natural enemies, human health or the environment. The interest of these parasitoids in biocontrol is evident because they kill the pest at the most critical stage (the egg) before the damage occurs. Furthermore, the low cost of production has encouraged the commercialisation of rearing *Trichogramma*.

At GMP, *Trichogramma* cards are brought to the field and release only at the hedge on the field at the rate of 12000 *Trichogramma*/ha which is far not enough to be effective. *Trichogramma* cards are well stapled on top leaves and the leave is bended to protect the cards from heat and heavy rain. In addition **grease is applied to protect the *Trichogramma* from predatory insects** (ants and spiders essentially) which is remarkable.

However the overall impression is that the releases are neither organized on the basis of borer damage levels nor at the best time of crop growth. In clear words there is no real release strategy taking into account the age of the field, the infestation levels and the variety.

Recommendations:

- 1) *Trichogramma* releases should be planned on a field information basis: age of the field, variety planted, damage levels from the previous year...*Trichogramma* production should be adapted accordingly.
- 2) Release should be done early in the morning between 7 to 9 am to avoid the heat. Release rates should be at least 100.000 *Trichogramma*/ha which is the recommended number in many countries. However, a production of 10 times of what is produced today is a real challenge and will require a dramatic improvement of *Corcyra* production.
- 3) *Trichogramma* cards should have less parasitized eggs (1000) to allow a better distribution of *Trichogramma* in the field (*Trichogramma* is not a highly mobile insect as opposed to *Cotesia*). Therefore we advise whenever possible to apply 100 cards of 1000 per ha if possible.
- 4) Release period should be between 1 month and 4 months, which is the usual oviposition period for stemborers. All fields of this age between June and September will be highly exposed to high damage.

3.3. *Cotesia flavipes*: rearing and release techniques

This larval parasitoid is widely used to control moth borers around the world in different agrosystems, including sugarcane. Brazil has succeeded to control his major stalk borer *Diatraea saccharalis* thanks to the use of *Cotesia flavipes*.

At GMP, *C. flavipes* is reared on *Chilo auricilius* because this host is easier to produce than *C. sacchariphagus*. However the general production in laboratory remains at a small scale and the rearing conditions, as for the other species, are not optimal. The larvae are reared in glass tubes and the humidity inside can be a concern. From our visit, there is a need to separate the kitchen devoted for making the artificial diet, to the other activities such as insect storage, insect manipulation, etc.

We think that the number of individuals produced and released at GMP to parasitize borer larvae is far too low (280 individuals per ha). In fact, in Brazil and Thailand, 2 big users of this parasitoid, they mass produce and release 2500 to 6000 *Cotesia* adult per hectare in 2 or 3 times! The rate used by GMP is certainly interesting to maintain a certain number of parasitoids in the field but cannot be seen as a sustainable strategy to reduce borer infestations. In addition, the effect of these releases on the borer population is not known, as no assessment is done after the releases.

Apart from a low dose, the timing of release doesn't even seem to be appropriate. In fact, the best time to release *Cotesia* is between 3 months to 6-7 months of cane growth simply because the overall peak of borer larvae occurs at this time. The objective of releasing this parasitoid should be to target the larval stage in the field and this is also the stage where sugarcane is in growing period and the chances to parasitize the larva is maximized. Therefore it is recommended here that the production and the releases are planned accordingly. This is the same logical frame that is considered for *Trichogramma* where the releases need to be done at a certain time.

Recommendations:

- 1) Specific room should be devoted to the rearing of *C. auricilius* and *C. flavipes* with all necessary equipment and conditions for good rearing and storage (see recommendations for *Corcyra* and *Trichogramma*)
- 2) Use right rate at the right time! This mean at least 2000 to 2500 *Cotesia flavipes* wich is almost 10 times more than the current rate.
- 3) Parasitoids should be released at different locations of the fields to make sure the dispersion and then the parasitism will be efficient, even if *Cotesia* is a good flyer and is able to search for host quite far from the release point.

3.4. Other aspects of IPM

Light traps

GMP has a unique grid of 8 light traps located in different sugarcane zones. It is certainly a good tool to monitor the moth borer populations of different species. We have suggested that the data be used to monitor the borer population (a weekly

survey would be ideal) all year round as an indicator to trigger biological control or any other pest management strategy.

Bat houses

Several 'Bat-houses' have been set up at the edge of sugarcane fields for conservation of insectivorous bats. It is known that the bats provide regulating services in pest management as they feed from insect pests such as white grubs (beetles), moth borers, crickets, armyworms, termites. Bats are also used in South Africa for pest control and conservation purposes in the sugarcane industry. At GMP, the research department is currently carrying out studies on bat populations and feeding habits (analysis of insect fragments in bat feces) with encouraging results.

CONCLUSION:

The visit has been very useful and we hope that this make a start for a sustainable collaboration between CIRAD entomologists and GMP. Despite an interesting biocontrol strategy, this company should improve the overall conditions for insect production in the laboratory and release techniques in the field. We think that this could be achieved step by step by putting more investments in order to renovate rooms and buy small equipments and air conditioning systems. In addition there is a real need for an assessment system of all pest management techniques, particularly for biological control which is the main component of IPM at GMP. We propose in the second part of this report a series of proposals to undertake this task.

It could be also helpful to investigate crop loss due to stem and top borers to assess the economic impact on sugar and cane yield (biomass). It is even possible to determine an economic injury level (EIL) to decide whether a pest control strategy should be implemented or not, based on the cost of interventions and the market value of the products at GMP (cane sugar, cane biomass). This will facilitate decision makings and the proposal 4 in this report can help to achieve this objective.

Proposal No.1 (2012-2013)

PARASITISM LEVELS OF SUGARCANE MOTH BORERS AT GMP

Location/site: Field/areas to be selected by GMP

This protocol will bring additional and useful information to the current release strategy of insect parasitoids in the field.

Objective:

- Determine parasitism levels (eggs, larvae, pupae) of sugarcane borers during the crop cycle
- Collect and identify parasitoid species in sugarcane fields

Period and Frequency:

The observations can start on young fields (age of 2 months) and stop when field is 10 months old, which corresponds to 8 months of sampling, or 32 weeks.

Sampling for borer eggs and larvae (and other key predators if found) should be done once a week on Monday (or Friday) on a designated sugarcane area/block (to be selected by the field team). The total time for searching to be completed should be 4 hours (e.g. from 8 am to noon). If the minimum number of samples required (= 10 larvae and 5 egg batches) is not reached, the search can be extended to a larger area (= additional time) the same day or the day after, depending on the availability of the field staff.

Teams

2 teams should be involved for the collection of samples: one in the field searching for eggs (F1), one in the field searching for larvae (F2). Note that the teams can help each other, in case one finishes before the other. We think that a good estimation of *Chilo* and *Scirpophaga* parasitism can be obtained if 30 larvae and 10 egg batches are collected per week.

1) Field procedures:

2 adjacent fields of 1ha (or more) with commercial varieties are needed to do the survey. This is to allow enough samples each week. If the eggs or larvae are not found in sufficient number, the additional eggs can be searched on field F2. Same thing for larvae.

- **Field 1 “eggs”**: Field devoted for egg collection. It is advised to divide the field into 4 quadrats to make sure that all parts of the field will be visited. Maximum 1 hour should be required for the inspection of each quadrat. For each quadrat, rows of sugarcane should be carefully inspected by one inspector from the border to the centre of the field by walking through slowly (see figure).

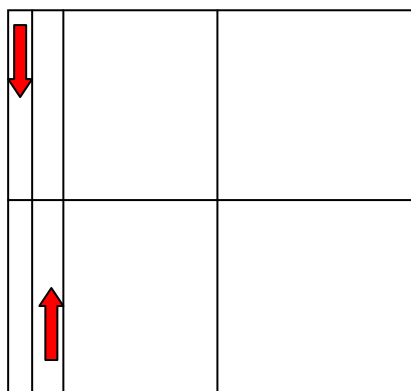
Careful examination of green leaves (from the bottom to the top, both sides of the leaf) of each sugarcane plant can be done for detecting the presence of *Chilo* eggs (egg batches form in 2 rows, white in colour if freshly parasitized, black if parasitized) and *Scirpophaga* eggs (covered by hairs). All egg batches should be cut out from the leaves and then placed in jar or vials (one egg batch per vial) and brought back to the

laboratory for further observations. All samples should be labelled (day of collection...). Remember that the objective here is to find a minimum 5 egg batches for each group (5 *Chilo* eggs, 5 *Scirpophaga* eggs regardless of the quadrat) bearing in mind that the ideal is 10.

- Field 2 “larvae/pupae”: Follow the same procedures as above.

Each quadrat should be carefully inspected row by row to search for plant having signs of attack and larvae have to be recovered by dissecting the young plant or stalk. Remember that a minimum of 10 larvae of each species has to be found, knowing that the ideal number for parasitism estimation is 30 larvae per borer species (If too complicated *Chilo sacchariphagus* and *C. auricilius* will be considered as one species). If different stages are found (including pupae), they should be kept separately in vials or plastic boxes with pieces of sugarcane: small (S), medium (M), Large (L), Pupae (P). If a larva is found dead/parasitized, it should be also collected and taken into account for further parasitism assessment. If a white cocoon (*Cotesia* spp.) is found next to a dead larva it will be also collected.

All samples collected should be kept in laboratory for further parasitism examination.



2. Laboratory Procedures

EGGS

Once the egg batches have been brought to the laboratory, all vials should be kept for observation for a maximum of 14 days. All egg batches (parasitized and supposedly unparasitized) should be counted for eggs and the number of eggs per batch be then determined.

The percent parasitism is calculated as the No. of eggs parasitized /number of eggs at the time of the collection. All egg parasitoids found should be kept in alcohol (80 to 90%) for further identification.

Chilo spp.

- For the white or yellow-orange egg batches (*Chilo* spp): egg batches should be checked every day until the neonates have hatched. Neonates should be counted and % of hatching determined.
- For the grey-black egg batches: all eggs can be observed under a microscope to separate eggs parasitized from egg with exit holes (parasitoids already emerged). For those without exit holes, batches should be observed every day until *Trichogramma* adults emerge. They should be counted and sex ratio

determined under a microscope (male can be distinguished from the female by looking at the antennae: Antennae of male *Trichogramma* have many long hairs. Antennae of females have a few short hairs.

Scirpophaga excerptalis

Same procedures as above. However *Scirpophaga* eggs are more difficult to count because of the hairy cover. Egg number can be estimated based on the number of larvae hatched. Parasitoids (*Trichogramma* spp., *Telenomus* spp. or other species) should be count per egg mass, labelled and kept in alcohol for further identification.

LARVAE AND PUPAE

Once the samples have been brought to the laboratory, the different stages should be counted (small, medium, large, pupae) and each individual should be kept in separate boxes on a piece of sugarcane for a day to day observation of parasitism. The percent parasitism is calculated as the No. of larvae parasitized /No. of larvae at the time of the collection.

LARVAE

Parasitism by *Cotesia flavipes* (Hymenoptera, Braconidae)

The gregarious endoparasitoid *C. flavipes* has a short lifespan of a few days and an initial eggload of 150 to 200 eggs. A female of this parasitoid allocates around 50 eggs in a host and the highest reproductive success is on later larval instars (4-6th) of borers. The egg to adult development time is 20 days and the sex ratio is usually female biased (60-70%). When parasitized a borer larva doesn't move a lot and doesn't feed. All *Cotesia* larvae come out from the body of the borer larvae and then they form a white cocoon to start pupating. Adults are black and small (2 mm long) and easy to recognise.

Other larval parasitoids

Elasmus zehntneri (Hymenoptera, Eulophidae) is an ectolarval parasitoid of *Scirpophaga*: The host range of this species appears to be limited to mature larvae of the top borer. Female deposit 25 to 30 eggs per host and 2 weeks are generally required to see the adults emerging from their cocoons.

Stenobracon deesae (Hymenoptera, Braconidae): solitary ectoparasite of stemborers.

tachinid flies: *Diatraeophaga striatalis*

PUPAE

Parasitoids:

Isotima javensis (Hymenoptera, Ichneumonidae)

Xanthopimpla xeminator (Hymenoptera, Ichneumonidae),

Tetrastichus spp., *Pediobius furvus* and *Trichospilus*.

3. Identification of parasitoids

Specimens kept in alcohol should be processed for morphological and DNA analysis. Taxonomy of parasitoids should be handled by specialists in Europe.

Nota: All datasheets, materials and field sites should be prepared before the collection. All data information is entered in datasheets for further analysis.

Proposal 2 for 2012/2013

Monitoring of sugarcane moth borers using synthetic pheromones and study on mating disruption for pest management purpose.

Aim:

- To monitor male populations of moth borers using synthetic pheromones during the crop cycle as a warning system for borer management decision
- Do preliminary tests on mating disruption (*) for pest management purpose.

(*) *The general effect of mating disruption is to confuse the male insects by masking the natural pheromone plumes, causing the males to follow “false pheromone trails” at the expense of finding mates, and affecting the males’ ability to respond to “calling” females. Consequently, the male population experiences a reduced probability of successfully locating and mating with females, which leads to the eventual cessation of breeding and collapse of the insect infestation. The [United States Environmental Protection Agency](#) considers mating disruption to be among the most environmentally friendly treatments used to reduce pest infestations. Mating disruption works best if large areas are treated with pheromones. Ten acres is a good minimum size for a successful MD program but larger areas are preferable.*

Justification:

Pheromones and semiochemicals are compounds produced naturally by insects that govern all aspects of their behaviour, including mating, aggregation, defence, host recognition and resource location. In recent years, pheromone-based pest management programs have been increasingly used to provide environmentally friendly approaches to control major agricultural pests, from mating disruption to mass trapping with “lure and kill”. Pheromone-baited traps are useful devices for monitoring moth population levels of stem borers (Kfir *et al.*, 2002). Trap catches of male moths can provide useful information for the timing of insecticide applications, releases of parasitoids and yield loss predictions (Van Rensburg, 1997, Wyatt, 1998). Campion & Nesbitt (1983) reviewed the progress in the identification and the utilization of sex pheromones for stem borer monitoring and concluded that mass trapping is unlikely to provide satisfactory control but that mating disruption is more likely to be effective. Synthetic pheromone blends for *Chilo suppressalis*, *C. sacchariphagus*, *Chilo indicus*, *Chilo auricilius* and *C. zacconius* have shown satisfactory attractiveness to male moths in the field (Beever *et al.*, 1990). The synthetic pheromones of the main 2 moth borers, *Chilo sacchariphagus* and *Scirpophaga excerptalis* are commercially available and have already been tested in field conditions in South-Africa, Reunion and India with good results (Way *et al.*, 2004; Mukunthan & Singaravelu, 2005). Mating disruption has been successfully used to control another borer species, *Scirpophaga incertulas* (yellow stem borer) with significant damage reduction in rice fields of India (Cork *et al.*, 1996).

Methodology

- Set up a grid of 8 pheromone traps in 2 sites/fields with medium to high levels of damage (sites to be selected). Traps should be placed at least 50 meters from each other to avoid pheromone interferences.
 - Each trap to be checked once a week
 - Pheromone vials to be changed every 4 weeks. Sticky papers to be cleaned and replaced
 - Count the number of moth borer (males) caught in the traps each week
 - According to borer species, different Trap designs (see Funnel and Delta traps) should be compared.
 - The effects of trap colour, trap spacing, trap height (1 and 2 m) and the type of pheromone dispenser and loading on catches should be also investigated.
- Evaluate the possibility of using pheromones for mating disruption (one trial to set up in a selected field at GMP). Traps should be loaded with vials at high concentration of blended pheromones. Study to focus on *Chilo sacchariphagus* and *Scirpophaga excerptalis*.
- Chilo and Scirpophaga pheromone vials can be purchased in UK, University, University of Greewic (contact: Dudley Farman, d.i.farman@gre.ac.uk)

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Type of pheromone traps that can be used:

1) Funnel Traps (AgriSense BCS)



Premium long lasting quality, all green colour, smooth exterior profile ; Bayonet fit, accept pheromone vials direct on lure holder or lures in a cage

2) Delta traps.



White delta traps with sticky liners from insect science in S.A. worked wonderfully well for me in Reunion when I was testing a new consignment of pheromone that I got from Dudley. I now use them for the S.A./moz. border grid.



Proposal 3 (2012-2013)

Assessing parasitism level of *Trichogramma spp.* for quality control in laboratory and field conditions.

Aim

To conduct control tests of egg parasitism from *Trichogramma chilonis* and *T. Japonicum* on the main targeted moth borers: *Chilo sacchariphagus* and *Scirpophaga excerptalis* in order to ascertain their effectiveness not only in the laboratory (quality control of the species) but in the fields during release operations.

Justification

Trichogramma spp. is widely used a biological agent to control stem and top borers. Despite these species have proven their efficacy there is a need to constantly check their ability to parasitize their original hosts, namely *Scirpophaga excerptalis* and *Chilo spp.* It is known that the capacity of parasitism can be reduced after years of production on a factitious host, *Corcyra cephalonica* (rice moth). The use of these hosts for the production of parasitoids instead of natural hosts is explained by the low cost and easiness of breeding and manipulation. However it implies that quality tests should be conducted all year round to guarantee the effectiveness of released parasitoids from the factories. Another justification of this proposal is that the parasitism level by *T. japonicum* on *Scirpophaga* is suspected low and therefore need to be carefully checked on regular basis.

Methodology

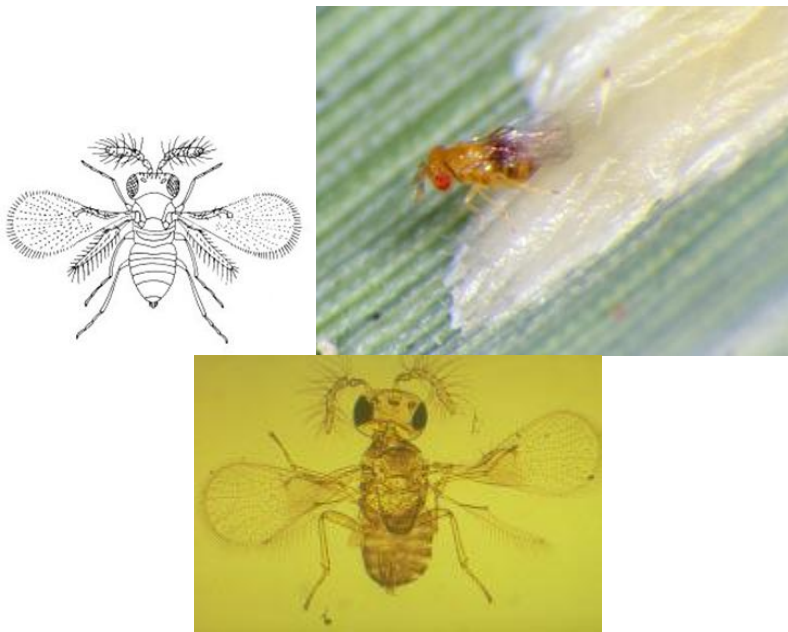
- Each month, collect fresh egg batches of *C. Sacchariphagus* and *S. excerptalis* from the fields (at least 10 per species), or alternatively from a colony established in shade house or laboratory.
- Expose the egg batches found (number will depend on field collection) once a month to *Trichogramma* species from *Corcyra* cards to check out for parasitism:
 1. in the laboratory (ex for : 10 females (aged 24 hours) are confined individually in glass tubes. One egg batch of each species (20 to 40 eggs) is introduced per tube/per female (a small droplet of honey is also provided). Borer eggs are removed from the tubes after 3 days of exposure and kept for the incubation period in separate vials. This operation is done once a month.
 2. in the field: egg batches of *Chilo* and/or *Scirpophaga* are stapled at the proximity of a tricho card (on the same stalk) released at the same time. Borer eggs are removed from the field after 3 days of exposure, brought to the lab for incubation in a vial/tube. Grease will be applied to protect egg batches from predation. This operation is done once a month.

Assessment includes:

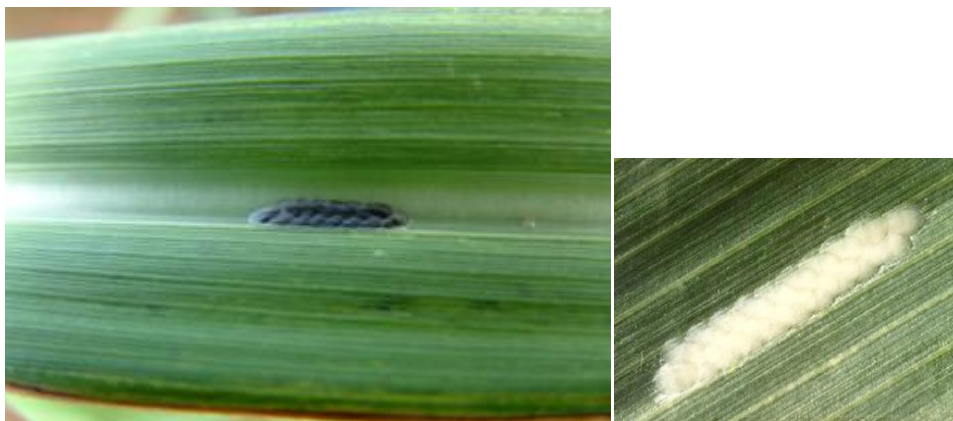
- Number of batches parasitised and eggs parasitised (black eggs)
- Number of individuals emerged from the eggs (fecundity)
- Longevity and sex-ratio of *Trichogramma* adults newly emerged

All the data are to be recorded in datasheets prepared in advance. Data and analyses will be conducted in collaboration with R.Goebel (CIRAD)

SOME IMAGES OF TRICHOGRAMMA SPP AND PARASITISM IN THE FIELD



Trichogramma under a microscope and in position to insert eggs in a *Chilo* egg batch (Right).



Black eggs of *Chilo sacchariphagus* = parasitized by *Trichogramma* (left) and white eggs (unparasitized, right)

PROPOSAL 4 (2012-2013)

Crop Loss due to moth borer and *Trichogramma* efficacy

(July/august 2012 to July-august 2013)

Aim of the field trial

- Assess actual and potential efficacy of *Trichogramma* biocontrol program
- Measure crop loss caused by stem borers (*Chilo sacchariphagus* and *C. auricilius*) and the top borer (*Scirpophaga excerptalis*).
- Use the trial as a demonstration experiment for the R&D people and grower visitors.

Location: Gunung Madu Plantation (GMP), Sumatra

Trial design

4 treatments (T):

- T1: *Trichogramma* released according to GMP procedure (1,5 to 4 months; weekly releases, 12000 trichogramma per ha the first week (8 cards of 1500 Tr) then 9000 per week during 7 weeks (6 cards of 1500 Tr). It means for 500 m² we should release 60 0 Tr for first week and 450 Tr during 7 weeks.
- T2 : *Trichogramma* released according to recommended procedure = 14 releases commencing 30 days after harvest (DAH) and finishing 120 DAH, 100.000 Trichogramma per ha and per week. This means for 500m² we release 5000 Trichogramma for each week.
- T3: Full protection with an Insecticide (a.i. Chlorantraniliprol 100 gr/l and Lambda Cyalothrin 50 gr/l) 2 ml/liter applied every 2 weeks from 1-8 months with a knapsack (15 or 20L).
- T4 : UTC (untreated control)
- Variety to be used : A susceptible GMP variety (to be discussed)

The trial has 5 replicates

Plot size 500 m² with 20 m of cane buffer between them

Observations/assessments (GMP staff)

- Damage at 3, 6, 9 months and just before harvest (12 months): 50 consecutive stalks x 4 (= 200 stalks per plot): number of stalk damaged, internodes damaged, length of internal borings, species and number of eggs, larvae and pupae found.
- Crop yield (cane weight) : estimated on the 4 middle rows (to be discussed)

- Sugar yield components measured (quantity and quality parameters): Brix, Pol, Purity, Fiber% cane and recoverable sugar (CCS) at harvest on 20 stalk samples

Relationship between damage and yields (biomass)

Observations/assessments at harvest on 50 stalks x 2 samples = 100 stalks per plots/treatment to be taken in the middle of each plot.

The step will be the following once in the field:

STEP 1 : stalk height, diameter and weight

- 1) Select 50 x 2 rows (100 stalks) in the middle of the plot
- 2) Each stalk will be detashed and numbered (use a marker) from 1 to 50 for each of the 2 groups/rows (2 x 50) per plot = 2000 stalks for 4 plots x 5 reps.
- 3) Cut each stalk at the bottom (very close to the ground) and the top of leaves (at the terminal visual dulap = last visible mark)
- 4) Weight of each marked stalk with the help of a portable scale
- 5) Measure the diameter (mm) of each stalk (2 measures = bottom and middle) with a Vernier calliper equipped with a small graduated rule.
- 6) Measure the height of each cane (to the Terminal Visual Dulap (TVD), the last purple strip/mark on the top).

STEP 2 : damage counts, intensity index of damage, insect collection

- 7) Count the visible internodes from the bottom (internode 1) to the top (internode X) and count damaged internodes (presence of holes) of each stalk.
- 8) Position of the damage along the stalk : presence of the damage at internode X or Y (ex : there are 2 holes at the internode 5 and one at the internode 15)
- 9) Collect borer larvae by cutting around the damaged internodes and estimate the stage (small – medium – large larvae, pupae)
- 10) Collect parasitoids (cocoon of *Cotesia* or pupae of *Diatraeophaga*) if any
- 11) slice each marked cane transversally
- 12) measure the length of damage inside the stalk for each internode

Material needed: Vernier caliper (diameter), knives, stick (measure height), indelible markers, excel data sheet of 50 lines (represent 50 stalks) and 25 columns (represent internodes) (total for the trial = 40 sheets) and scale for stalk weight.

Trichogramma spp. FIELD TRIAL LAYOUT (4.5 Ha)

